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Sensitivity of quinoa cv. 'Titicaca' to low salinity conditions

María del Carmen Rodríguez-Hernández[1,], Luna Morcillo[2], Idoia Garmendia[1]*

[1] Department of Environmental and Earth Sciences, University of Alicante, PO Box 99, 03080 Alicante, Spain
[2] Fundación Centro de Estudios Ambientales del Mediterráneo (CEAM), Joint Research Unit University of Alicante-CEAM, University of Alicante, PO Box 99, 03080 Alicante, Spain

ABSTRACT

Quinoa (*Chenopodium quinoa* Will.) is an annual herbaceous Andean plant. In recent years there is a growing interest on it due to its high quality as food, its wide adaptation to agroecological conditions and resistance to different abiotic stresses. In this work, we evaluate the growth pattern of quinoa plants cv. 'Titicaca', subjected to different levels of salinity, focusing on leaf production and nutrient content. In this sense, the results have shown that a high concentration of salinity negatively affects the growth of quinoa plants. In fact, plants grown with 200 mM NaCl reduced the photosynthetic rate and levels of chlorophylls and carotenoids in comparison with the rest of the treatments. Likewise, it has been proven how the progressive increase in salinity has negative effects on transpiration, stomatal conductance and photosynthetic rate, with significant subsequent reductions in shoot biomass, leaf area and nutrient adquisition, but without a decline in leaf dry weight (DW) production. However, the treatment of 200 mM NaCl demonstrated the best results regarding the water-use efficiency, as well as the number of saline glands. According to our results, the quinoa plant cv. 'Titicaca' seems to be tolerant to moderate concentrations of salinity (50–100 mM NaCl). This study could serve as a reference on this little known and cultivated species in the Mediterranean region, since it could become an alternative crop in areas with moderate salinity problems.

Keywords: biomass production, *Chenopodium quinoa*, photosynthetic rate, salt glands, salt stress, water use efficiency

INTRODUCTION

Environmental changes in the last decades have escalated to critical levels, presenting environmental risks to people, especially in terms of food supply, as it affects crop yield, production and quality (Alexandratos, 2005). Soil salinisation is one of the major environmental issues affecting crop production, especially in marginal landscapes or areas with limited resources (Rengasamy, 2010; Munns, 2011; Hussin et al., 2013). The intensive use of valuable natural resources such as land and water, along with high soil evapotranspiration and inefficient irrigation systems associated with poor water and soil management, inevitably accelerate secondary salinisation that usually results in the loss of productive areas (Munns, 2005; Hussin et al., 2013). In fact, soil and water salinity are wide-spread and currently, one-fifth of irrigated lands are salt-affected and 1.5 million hectares of lands are becoming unsuitable for agricultural production every year because of high salinity levels (Hossain, 2019).

In this sense, the Mediterranean region is characterised by an extremely variable climate with dry-hot summers and wet-cold winters (Ceccarelli et al., 2007). Crop production in the southern Mediterranean region is restricted by limited water resources, drought and salinity. The adaptation of agriculture to changing climatic conditions includes the use of suitable crops, for example, species or genotypes with improved tolerance to abiotic stresses, such as cold, drought or saline soils

*Corresponding author.
e-mail: maricarmen.rodriguez@ua.es (María del Carmen Rodríguez-Hernández).

(Rodríguez-Hernández et al., 2013; Fernández-García et al., 2014; Scheben et al., 2016; Cordero et al., 2019). One such crop, and perhaps the most promising, is quinoa. Quinoa (*Chenopodium quinoa* Willd.) is a native grain from the Andean highlands in South America that belongs to the Amaranthaceae family. It could be an excellent alternative crop in many regions of the world as is one of the few crops, if not the only one, that is capable of being successfully cultivated in the most extreme environmental conditions (Jacobsen et al., 2003). According to Adolf et al. (2013), quinoa is a halophyte that has the potential to become an important crop in arid regions and saline habitats and has also expanded in many parts of the world to satisfy new market niches for gluten-free foods. In general, quinoa combines a high inherent tolerance to salinity (Pulvento et al., 2012) and to other several environmental stress factors (Jacobsen et al., 2003; Razzaghi et al., 2011) together with a high nutritional quality (Repo-Carrasco et al., 2003). As a consequence, the demand for quinoa seeds has significantly increased over the last decade (Bazile et al., 2016). However, its production in the traditional areas of cultivation is not adequate to satisfy the increasing global demand, and thus it could be cultivated outside the equatorial Andes region of South America (Jacobsen, 2017).

Quinoa seeds are commonly known for their high nutritional value, mainly due to the amount of dietary fiber (7–10%), their high protein content and a wide range of minerals (Ca, Fe, Zn, Cu and Mn) and vitamins (B, C, and E), as well as health-promoting compounds, such as polyphenols (Ruales and Nair, 1993; Repo-Carrasco et al., 2003; Ruiz et al., 2014). On the other hand, quinoa leaves are considered nutritious vegetables, and based on their dry weight (DW), they present a better profile than grains. The protein content in their fresh leaves (14.77%) is greater than that in spinach (2.86%), chard (1.82%) and broccoli (2.98%) (Vazquez-Luna et al., 2019). Consequent to this large agricultural and nutritional potential, interest in quinoa has increased in recent years. Quinoa is traditionally classified into five ecotypes (highland, inter-Andean valley, salares, yungas and coastal lowlands), which are associated with the dispersion cores located in southern Peru and Bolivia close to Titicaca lake (Tapia, 2015).

During the last two decades, numerous articles in the literature have addressed salt and drought tolerance in quinoa (Jensen et al., 2000; Bhargava et al., 2006; Ruffino et al., 2010; Ruiz et al., 2014; Choukr-Allah et al., 2016). However, little is known about the growth of the 'Titicaca' cultivar (highland ecotype) in the Mediterranean region. 'Titicaca' cultivar represents one of the nine registered European quinoa cultivars (Jacobsen, 2017). In addition, it has properties that make it interesting as a result of its high antioxidant activity and its short time to flowering and to maturity compared to other cultivars (Reguera et al., 2018). According to Bazile et al. (2016), 'Titicaca' cultivar has a very good yield compared to other varieties; however, it presents a very heterogeneous yield and needs to be tested in more diverse conditions, considering its good yield production. On the other hand, there are an inadequacy in the number of studies that analyse the behaviour of this cultivar under greenhouse conditions and hydroponics, focusing on leaf production and nutritional content. In this sense, 'Titicaca' cultivar could be appropriate to leaf production as a product with high nutritional value. For this reason, this work aims to conduct more research into this cultivar of quinoa by analysing its behaviour against different levels of salinity, but this time under hydroponic and greenhouse conditions, and the impact of such salinity-levels on leaf biomass and mineral concentration. Thus, a broad study encompassing the biomass production parameters, as well as the water status, gas-exchange and quality parameters of this cultivar was assessed.

MATERIALS AND METHODS

Plant material and growth conditions

Seeds of quinoa cv. 'Titicaca' (provided by the Centre of Edafology and Applied Biology of Segura (CEBAS-CSIC) in Murcia, Spain) were disinfected with 0.5% NaClO during 2 h and pre-hydrated with aerated, deionised water for 22 h. Subsequently, they were germinated in vermiculite in a chamber with an air temperature (T) of 26/20°C day/night (D/N) and relative humidity (RH) of 60/80% D/N. The chamber light conditions were 16 h light–8 h dark cycle with a photosynthetically active radiation of 400 μmol · m^{-2} · s^{-1} that was supplied by a combination of fluorescent tubes (Philips TLD 36W/83 Germany and Silvana F36W/GRO, USA) and halogen lamps (Osram HQI-T 400W, Germany). After 10 days, seedlings were transferred to a greenhouse under semi-controlled conditions of T D/N: 25/18°C; RH D/N: 60/80% and received natural daylight (mean photosynthetic photon flux rate of 400 μmol · m^{-2} · s^{-1}). Seedlings were transplanted to 5 L plastic hydroponic containers with Hoagland areated nutrient solution (Hoagland and Arnon, 1938) and were divided into four homogeneous groups with 10 seedlings in each one. Four saline treatments were compared: control (0 mM), 50 mM, 100 mM and 200 mM of NaCl. After 7 days of salinity conditions, plants were harvested and the different determinations were performed. At the end of the experiment, the values for electrical conductivity (EC) of the nutrient solution from the non-saline container and the containes cultivated under 50 mM, 100 mM and 200 mM of NaCl were about 1.79 dS · m^{-1}, 3.21 dS · m^{-1}, 8.81 dS · m^{-1} 18.60 dS · m^{-1}, respectively.

Growth parameters

Plant DW was determined after drying at 80°C in an oven to a constant weight. In addition, shoot and root lengths were measured. Leaf area was calculated by using the app 'Easy Leaf Area Free' (Easlon and Bloom, 2014).

Water status

The leaf relative water content (RWC) was calculated according to Weatherley's (1950) method, using the following formula RWC (%): (FW − DW)/(TW − DW) × 100, where FW indicates fresh weight of the tissue, TW turgid weight and DW dry weight, respectively. This determination was performed in expanded young leaves collected at noon. The values for midday water potential of the stem and the root were measured by a Scholander pressure chamber (SKPM 1450/40, Skye Instruments Ltd., UK).

Photosynthetic pigments

The concentration of foliar photosynthetic pigments was determined in young leaves completely expanded and collected at noon, according to Séstak et al. (1971). The samples (20 mg FW) were included in 5 mL of 96% ethanol at 80°C for 10 min to extract the pigments. The absorbance of the extracts was spectrophotometrically measured and the equations reported by Lichtenthaler (1987) were used to calculate the concentration of chlorophylls and carotenoids.

Leaf gas exchange parameters

Leaf stomatal conductance (g_s), transpiration (E), photosynthetic rate (A) and instantaneous water use efficiency (WUE_{inst}) were measured using a portable photosynthesis system (CIRAS-3, PPSystem International, Amesbury, USA). The third fully-expanded leaf was chosen and the measurements were made early in the morning to ensure that we could obtain the highest values (Vilagrosa et al., 2003; Ahmadi et al., 2010).

Salt glands

The number of saline glands in the quinoa leaves, both in the adaxial and abaxial surface, were determined on three fresh young and fully-expanded leaves per plant. On each surface of the leaf, gland counts were made randomly over four visual fields of the sample using a microscope (Optech, B3 tri-eyepiece). The images were captured using a camera (IDS, UI-1460SE-C-HQ) and subsequently a representative area was selected to count the glands using the image-processing program ImageJ to analyse leaf surfaces (Schneider et al., 2012).

Mineral analysis

Leaf samples (0.5 g DW) were dry-ashed and dissolved in HCl according to Duque (1971). Potassium, magnesium, calcium, phosphorus, manganese, iron, zinc and sodium concentrations were determined using a Perkin Elmer Optima 4300 inductively coupled plasma optical emission spectroscopy (ICP-OES) (Perkin Elmer, USA). The operating parameters of the ICP-OES were the following: radio frequency power 1,300 W, nebulizer flow 0.85 L · min^{-1}, nebulizer pressure 30 psi, auxiliary gas flow 0.2 L · min^{-1}, sample introduction 1 mL · min^{-1}.

Statistics

The results were analysed with one-way analysis of variance (ANOVA) by the statistical program SPSS v.26. The means ± standard deviation (SD) were calculated and – when the F ratio was significant – least significant differences were evaluated by the Duncan test. Significance levels were always set at 5%.

RESULTS

Growth parameters

Growth parameters showed significant differences regarding to the salt treatment (Table 1). Shoot DW was significantly reduced with salinity treatment over 100 mM, while leaf DW maintained constancy even at high salt conditions. However, leaf area was negatively affected due to the application of 200 mM NaCl, as occurred with the parameter of shoot height with a reduction of 68% and 19%, respectively, comparing to control. Moreover, the results showed a significant decrease in both root DW and root length even with low salinity levels (50 mM).

Water status

With reference to leaf water status, plants grown at high salinity conditions (200 mM) had lower RWC than control plants (Table 2) with a reduction of 16.5% compared to control. Furthermore, the water potential of the stem and root decreased significantly as salinity level increased over 50 mM, with minimum values observed in plants subjected to 200 mM NaCl (−0.76 MPa and −0.30 MPa, respectively).

Table 1. Growth parameters of quinoa cv. 'Titicaca' subjected to different salt conditions. Means ($n = 10$) ± SD were compared with Duncan test.

Treatment	Shoot DW (g · $plant^{-1}$)	Leaf DW (g · $plant^{-1}$)	Root DW (g · $plant^{-1}$)	Leaf area (cm^2)	Shoot height (cm)	Root length (cm)
Control	2.04 ± 0.30 a	1.09 ± 0.27 a	0.47 ± 0.10 a	375.61 ± 60.67 a	24.75 ± 2.32 a	74.25 ± 8.99 a
50 mM	1.72 ± 0.31 ab	1.23 ± 0.43 a	0.37 ± 0.07 b	368.97 ± 68.47 a	23.25 ± 3.56 a	56.38 ± 7.79 b
100 mM	1.38 ± 0.12 bc	1.06 ± 0.37 a	0.37 ± 0.02 b	358.86 ± 31.38 a	25.36 ± 1.91 a	55.88 ± 7.26 b
200 mM	1.12 ± 0.11 c	0.94 ± 0.20 a	0.33 ± 0.06 b	120.36 ± 46.84 b	20.07 ± 0.73 b	41.13 ± 7.89 c

Within each column, values followed by a common letter are not significantly different ($p \leq 0.05$).

DW, dry weight; SD, standard deviation.

Photosynthetic pigments

In relation to the photosynthetic pigments (Table 3), the results indicated that only when plants were cultivated with 200 mM NaCl, both chlorophylls' and carotenoids' concentrations decreased to values of 15.45 mg · g^{-1} leaf DW and 2.57 mg · g^{-1} leaf DW, respectively, compared to the rest of the treatments, which had no differences between them.

Leaf gas exchange parameters

The results of gas exchange measurements (Figure 1) confirm that E and A, as well as g_s, were higher in the control treatment than the plants subjected to salinity, followed by the plants grown with 50 mM or 100 mM, and finally plants grown with 200 mM NaCl, a treatment which presented significantly lower values. On the contrary, the water-use efficiency values increased commenstruately with the increase in the severity of salinity.

Salt glands

Saline conditions induced salt gland development in leaves of quinoa cv. 'Titicaca' (Table 4). Number of salt glands, in both adaxial and abaxial surfaces of leaves, increased with NaCl concentration, showing significantly higher salt gland density in treatment with 200 mM NaCl compared to the control treatment. Furthermore, despite the fact that there were no significant differences between the two surfaces (adaxial and abaxial), a slight enhancement of gland number in the abaxial surface was observed.

Table 2. Leaf RWC and stem and root water potential (ψ) of quinoa cv. 'Titicaca' subjected to different salt conditions. Means (n = 5) ± SD were compared with Duncan test.

Treatment	RWC (%)	Stem ψ (MPa)	Root ψ (MPa)
Control	74.53 ± 5.52 a	−0.25 ± 0.05 a	−0.13 ± 0.05 a
50 mM	72.20 ± 6.18 a	−0.44 ± 0.05 b	−0.23 ± 0.05 b
100 mM	72.03 ± 7.74 a	−0.74 ± 0.18 c	−0.23 ± 0.05 b
200 mM	59.96 ± 2.90 b	−0.76 ± 0.05 c	−0.30 ± 0.07 c

Within each column, values followed by a common letter are not significantly different ($p \leq 0.05$).
RWC, relative water content; SD, standard deviation.

Table 3. Leaf photosynthetic pigment concentration of quinoa cv. 'Titicaca' subjected to different salt conditions. Means (n = 10) ± SD were compared with Duncan test.

Treatment	Photosynthetic pigments (mg · g^{-1} leaf DW)	
	Chlorophylls	Carotenoids
Control	23.42 ± 1.99 a	4.38 ± 0.40 a
50 mM	22.85 ± 0.97 a	4.31 ± 1.49 a
100 mM	22.68 ± 1.45 a	3.27 ± 0.76 ab
200 mM	15.45 ± 2.87 b	2.57 ± 0.47 b

Within each column, values followed by a common letter are not significantly different ($p \leq 0.05$).
DW, dry weight; SD, standard deviation.

Mineral analysis

The effect of salinity in leaf mineral concentration of quinoa cv. 'Titicaca' was also assessed (Table 5). Plants grown with 200 mM NaCl had significantly lower concentrations of the macronutrients Mg, K and P compared to the rest of treatments. On the other hand, the amount of Ca was lower as salt concentration increased over 50 mM. In respect of the micronutrients, high salinity levels caused a reduction in leaf Fe and Mn of 26% and 43%, respectively, compared to control. Nevertheless, Zn concentration had not a clear trend related to the effect of salinity. Finally, as expected, the accumulation of Na was higher as the amount of salt stress increased.

DISCUSSION

In saline environments, plant vulnerability is determined by the concentration and the specific ions involved; therefore, adaptation to this environment is crucial for the establishment and development of the species (Koyro and Eisa, 2008). In this sense, the wide genetic variability in salinity tolerance in quinoa provides an excellent source for selection and breeding for improved tolerance (Ruiz-Carrasco et al., 2011; Zurita-Silva et al., 2014). According to Hinojosa et al. (2018), quinoa can tolerate high levels of salinity, ranging from a salt concentration of 150–750 mM of NaCl (~15–75 dS · m^{-1}), which is greater than the salinity of seawater (>45 dS · m^{-1}). However, the optimal salinity conditions for quinoa growth are in the range of 100–200 mM NaCl (Eisa et al., 2017). Although quinoa has been classified as a highly salt-tolerant species (Jacobsen et al., 2003; Hariadi et al., 2011; Razzaghi et al., 2011; Eisa et al., 2012; Adolf et al., 2013), many quinoa cultivars show distinct variability in their germination and growth in response to salinity. Sun et al. (2014) revealed that 'Titicaca' cultivar is more sensitive to salinity than other quinoa varieties such as the Achachino variety. In our case, there was a significant decrease in shoot DW and root length of cv. 'Titicaca' from 100 mM of NaCl, and this decrease was more pronounced in the treatment with 200 mM (18.60 dS · m^{-1}). These results are in accordance with the previous work of Pacsi (2015), who observed that the INIA Salcedo variety did not show significant variations with low salinity concentrations, but that at concentrations of 200 mM the growth underwent a significant reduction.

Similarly, Gómez-Pando et al. (2010) found that some genotypes of quinoa showed a reduction in height when they were grown under salinity, as happened in our results with the 'Titicaca' cultivar, which exhibited a reduction in the length of shoots in plants grown with 200 mM NaCl and in the length of roots in plants grown with >50 mM NaCl. In addition, roots seemed to be more sensitive to salinity, as described by Panuccio et al. (2014), who demonstrated that quinoa is more susceptible to ion toxicity in roots than in above-ground organs. On

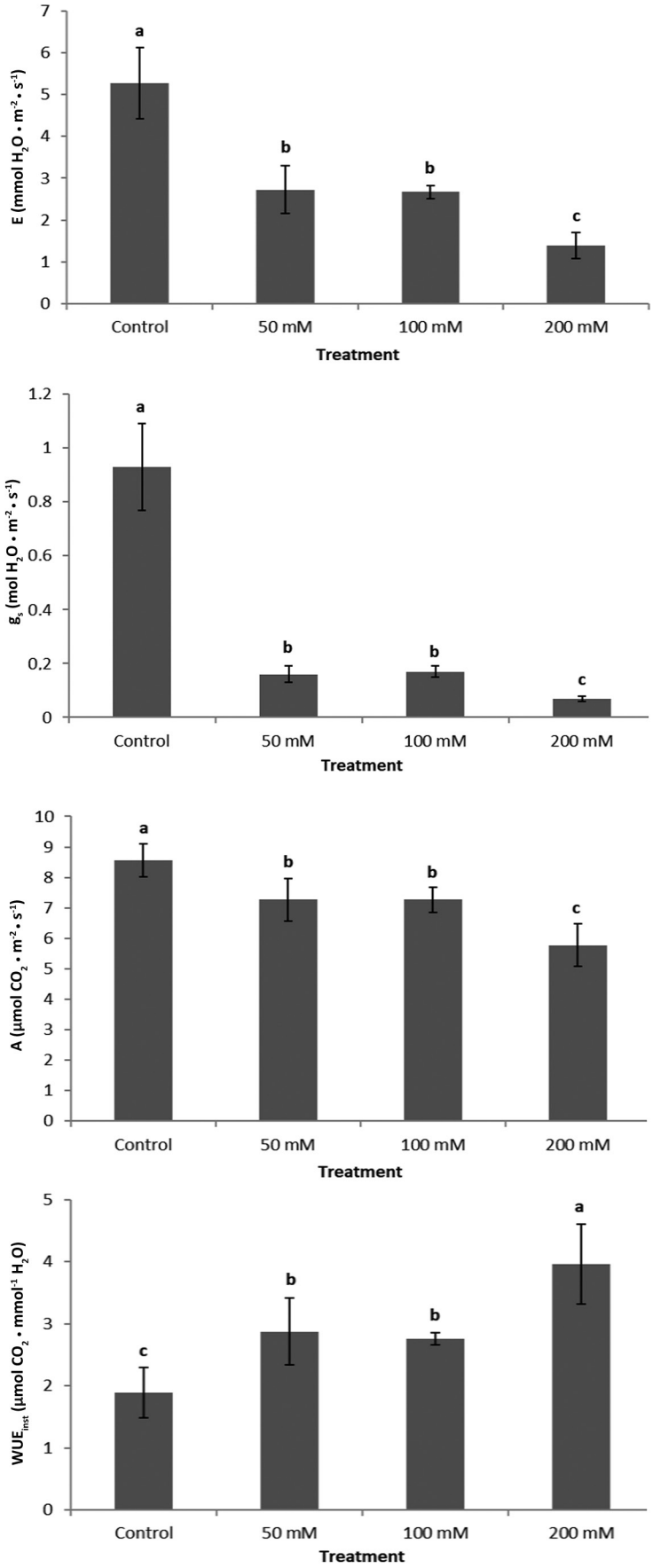

Figure 1. E, g_s, A and WUE_{inst} of quinoa cv. 'Titicaca' subjected to different salt conditions. Means ($n = 10$) ± SD were compared with Duncan test. Within each parameter, values followed by a common letter are not significantly different ($p \leq 0.05$). A, photosynthetic rate, E, leaf traspiration rate; g_s, stomatal conductance; WUE_{inst}, instantaneous water use efficiency; SD, standard deviation.

Table 4. Salt glands in leaves of quinoa cv. 'Titicaca' subjected to different salt conditions. Means (n = 5 plants, 15 leaves) ± SD were compared with Duncan test.

	Salt gland density (gland number · cm^{-2})	
Treatment	Adaxial surface	Abaxial surface
Control	339.08 ± 87.66 bx	368.90 ± 53.55 bx
50 mM	418.06 ± 100.64 abx	416.80 ± 91.44 abx
100 mM	427.27 ± 98.20 abx	477.45 ± 105.99 ax
200 mM	497.60 ± 132.24 ax	523.75 ± 151.20 ax

Within each column (a, b) or line (x), values followed by a common letter are not significantly different ($p \leq 0.05$).
SD, standard deviation.

the other hand, Ruiz-Carrasco et al. (2011), in studying 'BO78' quinoa variety, found a significant reduction in root length when the quinoa plants were subjected to 300 mM NaCl. In this work with 'Titicaca' cultivar, a significant decrease occurred with salinity levels over 50 mM, which indicates that this cultivar could be more sensitive to salinity. Furthermore, salinity caused a lower concentration of minerals such as K, Mg, Ca, P, Fe and Mn, which could be another factor that induced the decrease of the growth parameters (Amirjani, 2010). This plant biomass reduction, associated with saline conditions, can negatively affect seed yield of quinoa (Eisa et al., 2012; Talebnejad and Sepaskhah, 2015). Moreover, in accordance with Vazquez-Luna et al. (2019) and as stated above, quinoa leaves are considered nutritious vegetables. In this work, salinity conditions disminished mineral concentration of quinoa leaves, reducing its nutritional value.

A parameter widely used to characterise the plant's assimilation capacity is the leaf area (Hoogenboom et al., 1987). In this sense, as was seen in this work, 'Titicaca' cultivar showed a reduction in leaf area as salinity increased, a phenomenon that is similar to the one previously described by Hasegawa et al. (2000) and Koca et al. (2017). However, the leaf biomass was maintained with salinity levels below 200 mM, which could be related to the thickening of the leaves as a successful trait for plant species growing under saline conditions (Koca et al., 2017). Leaf thickening is considered as a mechanism to increase the water retention by mesophyll tissues to counteract salt toxicity (Naz et al., 2014).

Further, the reduction of the leaf area with salinity could indicate that salt excess has a negative impact on the quinoa's light absorption, and consequently, on its photosynthetic capacity. Salinity stress results in a decrease of photosynthesis in a wide variety of plant species (Sudhir and Murthy, 2004). However, many halophytes show a high rate of photosynthesis under elevated salinity conditions (Andersone et al., 2012), depending on the level of salt tolerance of the species and/or genotype (Brock et al., 2007). In this sense, Eisa et al. (2012) observed reduction of photosynthetic rate of quinoa with treatments above 100 mM NaCl, while in this work, cv. 'Titicaca' showed a slight but

Table 5. Foliar concentration of nutrients in quinoa cv. 'Titicaca' subjected to different salt conditions. Means (n = 5) ± SD were compared with Duncan test.

Treatment	Mg (mg · g^{-1} DW)	K (mg · g^{-1} DW)	P (mg · g^{-1} DW)	Ca (mg · g^{-1} DW)	Fe (µg · g^{-1} DW)	Mn (µg · g^{-1} DW)	Zn (µg · g^{-1} DW)	Na (mg · g^{-1} DW)
Control	9.23 ± 0.59 a	95.19 ± 6.04 a	5.79 ± 0.35 b	23.96 ± 1.49 a	72.24 ± 3.14 a	85.24 ± 12.88 a	90.37 ± 10.77 bc	1.52 ± 0.06 d
50 mM	9.10 ± 0.69 a	89.38 ± 13.24 ab	6.29 ± 0.18 ab	16.83 ± 1.56 b	71.40 ± 4.30 a	78.60 ± 2.59 a	106.59 ± 17.04 ab	29.86 ± 5.91 c
100 mM	8.62 ± 0.38 a	81.70 ± 4.52 bc	6.85 ± 0.89 a	13.14 ± 0.32 c	63.35 ± 9.25 ab	75.03 ± 2.08 a	129.35 ± 14.85 a	49.44 ± 9.72 b
200 mM	6.74 ± 0.36 b	69.47 ± 4.30 c	3.39 ± 0.12 c	10.90 ± 1.04 d	53.25 ± 5.67 b	48.33 ± 1.34 b	68.09 ± 10.46 c	63.26 ± 6.07 a

Within each column, values followed by a common letter are not significantly different ($p \leq 0.05$).
DW, dry weight; SD, standard deviation.

significant reduction even with 50 mM. Moreover, it is known that there is a positive correlation between the photosynthetic capacity and chlorophyll content (Morgan and LeCain, 1991). Our results, which were obtained in relation to the photosynthetic rate, agree with the levels of chlorophylls and carotenoids observed, showing a decreasing trend as the salinity concentration increases, similar to previous works (Lampkin, 1990; Adolf et al., 2012; Manaa et al., 2019). It is noteworthy to mention that salinity levels below 100 mM had similar pigment concentration than controls, unlike what was described by Eisa et al. (2012), where slight salinity levels (50 mM) stimulated pigment production.

The effects of the photosynthetic rate's decrease can be caused by diffusion limitations through stomatal closure and/or biochemical limitations, mainly to Rubisco, that affect the CO_2 assimilation rate (Flexas et al., 2016). The correlation between photosynthetic rate and g_s in different varieties of quinoa was demonstrated by González et al. (2014) and Gámez et al. (2019). Causes of the decreased photosynthesis in this work with quinoa cv. 'Titicaca' due to salinity could be diffusional, because its stomata closed from 50 mM NaCl treatment. Furthermore, it is known that one of the mechanisms of response to salinity in quinoa is to reduce water loss through fast stomatal closure (Eisa et al., 2012; Ruiz et al., 2016). ABA regulation seems to be one of the mechanisms utilised by quinoa, when facing salinity-inducing decrease of turgor of stomata guard cells and thus stomatal closure (Zang et al., 2006; Jacobsen et al., 2009). In this work, a decrease in transpiration rate was also observed. This is evident when an increase in the concentration of NaCl causes higher values of WUE (Cocozza et al., 2013; Sun et al., 2014; Miranda-Apodaca et al., 2018). In this sense, one aspect that deserves further comment is that in our work, the reduction in the transpiration rate (73.62%) induced by high concentration of salts was proportionally greater than the one of the photosynthetic rate (32.40%), which could improve WUE according to Eisa et al. (2012). Thus, low levels of salinity could be beneficial in terms of water consumption without negative effect in plant aboveground biomass.

It is known that salinity causes a reduction in leaf water potential (Razzaghi et al., 2011; Eisa et al., 2012), as this work demonstrates. Moreover, as observed by Munns (2009), root Ψ also suffered a significant decrease in saline treatment of 200 mM of NaCl, although in this work with cv. 'Titicaca', reductions were significant at 50 mM of NaCl. According to Adolf et al. (2013), decline of Ψis due to an osmotic adjustment carried out by quinoa under saline stress due to an accumulation of Na in vacuoles. In this sense, our results showed that the amount of Na was higher as the salinity increased, while the amount of K, Mg and Ca decreased with salinity consequent to being antagonistic to Na (Niu et al., 1995; Dodd et al., 2010). In addition, in terms of leaf water content, it was observed that quinoa remains unchanged until the treatment of 200 mM NaCl. Similar results were obtained by Riccardi et al. (2014), who observed that quinoa grown with 200 mM NaCl undergoes a significant reduction in water content due to a decrease in quinoa water absorption.

Saline glands seem to be an efficient mechanism to resist drought and salinity (Agarie et al., 2007; Ben-Hassine et al., 2009). Salt-secreting structures, namely salt bladders and salt glands, are the unique structures that directly secrete salt ions out of the plant (Shabala et al., 2014; Yuan et al., 2015), with most studies concentrating on two plants, *C. quinoa* and *Limonium bicolor*. Quinoa is a typical halophyte that possesses salt bladders, and its salt secretion mechanism and salt transport pathway were illustrated in detail by Shabala et al. (2014). Conversely to the results of Orsini et al. (2011) and Ruiz et al. (2016), we observed that salt exposure caused a significant enhancement in the number of saline glands in comparison with plants which were not exposed to salt treatment. On the other hand, there were no significant differences in the salt gland density among the saline treatments tested. In our cv. 'Titicaca' plants grown under hydroponic conditions, the exclusion mechanism based on saline glands could have reached saturation at low salinity levels. Orsini et al. (2011), working with the variety 'B078' of quinoa, found a saturation of this exclusion mechanism at 150 mM NaCl.

CONCLUSIONS

To conclude, quinoa cultivar 'Titicaca' is not as effective in tolerating salinity as expected. In fact, a salt condition of 100 mM that persisted for a week was severe enough to reduce the photosynthetic rate and the shoot biomass of plants. Despite that fact that the quinoa plant has been classified as a halophyte, genotype variations and understanding of the mechanisms of adaptation and tolerance against salinity are fundamental aspects to be taken into account in future research. In this sense, this study could serve as a foundational model for future studies on this peculiar species, the seeds and leaves of which are of high nutritional value.

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AUTHOR CONTRIBUTIONS

All authors contributed equally to research and manuscript-writing.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

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